Time-lapse studies on the effects of silver nanoparticles in the Caco-2/HT29 model of intestinal barrier

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The use of nanoparticles (NPs) has increased in the last years, particularly in food-grade products, where NPs are used to improve texture, flavor, microbiological control, food processing and packaging. In this context, silver nanoparticles (AgNPs) are highly used by the food-packaging and pharmacology industries due to their antimicrobial properties. Since the health effects of this constant exposure are not well-known, the assessment of potential risks for humans related to AgNPs intake became crucial.

The objective of this study is to analyze the effects of the exposure to AgNPs over the gastrointestinal epithelium (morphologically and functionally) over a period of 96 h. To this aim, we used an *in vitro* co-culture monolayer model composed of differentiated Caco-2 and HT29 cells, which respectively mimic enterocytes and goblet cells. We have evaluated the effects of two different sub-cytotoxic concentrations of AgNPs: the estimated daily intake of Ag of about 80 µg (2.58 µg/mL), and the highest non-cytotoxic concentration tested in our system (100 µg/mL). The evaluation of parameters such as the trans-epithelial electrical resistance (TEER) and paracellular permeability (LY), will give insight on the effects of AgNPs over the barrier's integrity. Confocal microscopy images will give us information on the uptake and cellular localization of the nanoparticles. Changes in the expression of intestinal epithelial markers by RT-PCR and a Western Blot, such as brush border and tight-junctions proteins, will give us further information on the possible changes regarding the monolayer's digestive and barrier functions. Finally, the quantification the translocation through will measure the ability of AgNPs to cross the intestinal barrier.